product[6] is provided by the action of lithium aluminum hydride on this intermediate V.[7]

Acknowledgment.—The authors are grateful to Dr. H. T. Openshaw for a sample of isoemetine hydrobromide. Financial support was provided by the research committee of the Graduate School.

---

[7] It is pertinent that in the case of the Δ^3-dehydroyohimbane (D-E *trans*) and similar (*cf.* ref. 4) systems, catalytic reduction also affords the more stable 3,15,20-*cis-trans* product (E. E. van Tamelen, M. Shamma and P. E. Aldrich, THIS JOURNAL, **78**, 4628 (1956).

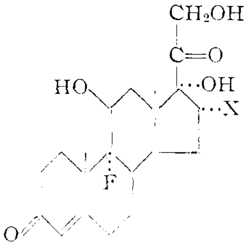
DEPARTMENT OF CHEMISTRY
UNIVERSITY OF WISCONSIN
MADISON, WISCONSIN
EUGENE E. VAN TAMELEN
PAUL E. ALDRICH
JACKSON B. HESTER, JR.

RECEIVED JUNE 17, 1957

OXIDATION OF STEROIDS BY MICROORGANISMS. IV. 16α-HYDROXYLATION OF 9α-FLUOROHYDROCORTISONE AND 9α-FLUOROPREDNISOLONE BY *Streptomyces roseochromogenus*

Sir:

The introduction of a 16α-hydroxyl group into 9α-fluorohydrocortisone (I) and 9α-fluoroprednisolone (II) has been shown by Bernstein, *et al.*,[1] to result in complete suppression of the salt-retaining properties of these steroids without appreciably impairing their glucocorticoid activity. Moreover, preliminary studies in man[2] have demonstrated the anti-arthritic activity of 9α-fluoro-16α-hydroxyprednisolone (triamcinolone) and have confirmed its lack of salt-retaining activity. An efficient synthesis of this complex steroid is therefore of considerable practical interest.



I, X = H
II, X = H; $\Delta^{1,2}$
III, X = OH
IV, X = OH; $\Delta^{1,2}$

Applying the microbiological 16α-hydroxylation reaction first reported from this laboratory[3,4] to I and II we have succeeded in preparing 16α-hydroxy-9α-fluorohydrocortisone (III) and 16α-hydroxy-9α-fluoroprednisolone (IV) in yields of 50% and 20%, respectively.

A fermentation medium containing soybean meal (30 g.), glucose (20 g.), soybean oil (4.4 g.) and calcium carbonate (0.050 g.) in distilled water (2 l.) was distributed over 40 250-ml. erlenmeyer flasks, steam-sterilized for 30 minutes at 120° and after addition of the steroid (1 g. dissolved or suspended in 40 ml. of methanol) inoculated with vegetative growth of *Streptomyces roseochromogenus*.[5] The flasks were incubated at 25° for 4 to 7 days with rotatory mechanical shaking at 280 r.p.m. in a 2 in. radius, the mycelium filtered off and washed with water. The filtrate was extracted with methyl isobutyl ketone (three 800-ml. portions), the extract concentrated to small volume, cooled and the resulting crystals recrystallized from ethyl alcohol. The properties of III (m.p. 250–252°, $[\alpha]_D$ +97° (*c* 0.99 in pyridine); $\lambda_{\max}^{\text{alc}}$ 238 mμ (15,000); $\lambda_{\max}^{\text{Nujol}}$ 2.79, 2.98, 5.82, 6.00, 6.17μ[6]; *Anal.* Found: C, 63.77; H, 7.32) and IV (m.p. 248–250°; $[\alpha]^{23}_D$ +71° (c 0.35 in acetone); $\lambda_{\max}^{\text{Nujol}}$ 2.95, 5.85, 6.02, 6.16, 6.24, 11.26 μ; diacetate: m.p. 170–180° (with gas evolution); $[\alpha]^{23}_D$ +28° (*c* 0.38 in $CHCl_3$); *Anal.* Found: C, 62.34; H, 6.74) are in harmony with those reported by Bernstein, *et al.*[1]

Oxidation of the diacetates of III and IV with chromic and sulfuric acids in acetone gave, respectively, the diacetates of 16α-hydroxy-9α-fluorocortisone, m.p. 215–217°; $[\alpha]^{23}_D$ +94° (*c* 0.35 in $CHCl_3$); $\lambda_{\max}^{\text{alc}}$ 235 mμ (15,100); $\lambda_{\max}^{\text{Nujol}}$ 2.94, 5.75, 6.00, 6.19 μ; *Anal.* Found: C, 63.03; H, 6.53, and of 16α-hydroxy-9α-fluoroprednisone, m.p. 204–206°; $[\alpha]^{23}_D$ +90° (c 0.41 in $CHCl_3$); $\lambda_{\max}^{\text{alc}}$ 235 mμ (16,200); $\lambda_{\max}^{\text{Nujol}}$ 2.97, 5.75, 5.99, 6.15, 6.22, 11.28 μ; *Anal.* Found: 63.13; H, 6.25.

The biosynthetic conversion of III into IV by means of *Corynebacterium simplex* has been reported.[1] We have performed this reaction in 65% yield with a strain of *Mycobacterium rhodochrous*[7] adding the steroid (0.5 g./*l.*) to a 24-hour old culture in a medium containing yeast extract, tryptone, pentane, glucose calcium, and incubating for five hours.

---

[1] S. Bernstein, R. H. Lenhard, W. S. Allen, M. Heller, R. Littell, S. M. Stolar, L. I. Feldman, and R. H. Blank, THIS JOURNAL, **78**, 5693 (1956).

[2] L. Hellman, B. Zumoff, M. K. Schwartz, T. F. Gallagher, C. A. Berntsen and R. H. Freyberg, Abstract of Papers presented at the 3rd Interim Meeting of the American Rheumatism Association, Bethesda, Md., Nov. 30, 1956.

[3] D. Perlman, E. O. Titus and J. Fried, THIS JOURNAL, **74**, 2126 (1952).

[4] J. Fried, R. W. Thoma, D. Perlman, J. E. Herz, and A. Borman, *Recent Progr. Hormone Research*, **9**, 149 (1955).

[5] Waksman Collection Number 3689, Rutgers University, New Brunswick, N. J.

[6] Occasionally a polymorphic form of III was obtained, which lacked the 2.79 μ band and showed an entirely different picture in the fingerprint region.

[7] This culture, isolated by Dr. J. O. Lampen of our laboratories, is identified as SC 2318 in our collection. With regard to its classification *cf.* R. Gordon and J. M. Mihm, *J. Bact.*, **73**, 15 (1957). Its dehydrogenating properties were discovered in these laboratories by Dr. H. Kroll.

THE SQUIBB INSTITUTE FOR
MEDICAL RESEARCH
NEW BRUNSWICK, N. J.
RICHARD W. THOMA
JOSEF FRIED
SALVATORE BONANNO
PAUL GRABOWICH

RECEIVED JULY 31, 1957

TOTAL SYNTHESIS OF 17-DESMETHOXYDESERPIDINE

Sir:

In a previous communication[1] we described the synthesis of the unsaturated lactone (I). We now wish to report the conversion of this intermediate to 17-desmethoxydeserpidine.

Reduction of the unsaturated lactone (I) with hydrogen and a highly active platinum catalyst for sixteen hours gave predominantly the saturated lactone (II), m.p. 267–269° (found: C, 74.81; H, 6.98; $\lambda_{\max}^{\text{Nujol}}$ 5.71 μ) as well as a small amount of an isomeric lactone (III), m.p. 307–309° (found: C, 74.25; H, 6.86; $\lambda_{\max}^{\text{Nujol}}$ 5.65 μ). When the product was allowed to remain in contact with the

---

[1] F. L. Weisenborn and H. E. Applegate, THIS JOURNAL, **78**, 2021 (1956).